

Vaccination Status and Antibody Titers against Rubella and Measles among Japanese Female College Students Majoring in Childcare between 2015 and 2018

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In 2014, for the protection of medical workers against measles and rubella infection, the Japanese Society for Infection Prevention and Control (JSIPC) recommended either maintaining antibody titers of seroprotective range or two-dose vaccination. JSIPC defined antibody titers into 3 ranges: seroprotective as expected prevention of infection, seronegative as under detection levels, and seropositive as antibody titers ranged between seronegative and seroprotective. This study aimed to explore the association between the number of vaccine doses received and the antibody titers against measles and rubella among Japanese college students majoring in childcare. A total of 841 female students with no history of measles or rubella were serologically screened at the time of college admission between 2015 and 2018. All 841 students had been vaccinated against measles; 738 (87.8%) received two doses of the measles vaccine and 103 (12.2%) received one dose. Likewise, 839 students, except for two, had been vaccinated against rubella; 719 (85.7%) received two doses of the rubella vaccine and 120 (14.3%) received one dose. We thus found that 107 students (12.7%) were seropositive for measles-specific IgG and 731 (86.9%) attained seroprotective titers. By contrast, in case of rubella-specific IgG, only 462 students (55.1%) attained seroprotective titers, and 371 students (44.1%) were seropositive. The two students without receiving rubella vaccination were classified as seronegative. In conclusion, despite that > 85% of students surveyed had received two doses of measles and rubella vaccines, a substantial number of students remain susceptible to measles and especially rubella at the time of college admission.

Keywords: immunization; measles; rubella; students majoring in childcare; vaccine

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Introduction

In an effort to eliminate measles and rubella, the Japanese government implemented a two-dose measles-rubella vaccination schedule during childhood in 2006 (IDSC, Infectious Disease Surveillance Center, NIID, National Institute of Infectious Diseases 2007) and a 5-year measles-rubella catch-up vaccination campaign conducted from 2008 to 2012 for cohorts aged 13 and 18 years (Kimura et al. 2013; IDSC, NIID 2015). In Japan, the person who was born after 2006 receives two-dose measles-rubella vaccination aged 1 year and 5 years, on the other hand, the person who was born between 1995-2005 received two-dose measles-rubella vaccination aged 1 year

and 13 years. Viral isolations and detections since November 2010 found that there have been only cases with imported viruses and there are no cases due to the genotype D5 that caused the epidemic in Japan from 2007 to 2008. In 2015, WHO, the World Health Organization verified that Japan had achieved measles elimination. However, there have been outbreaks spreading from imported case. (WHO 2015; IDSC, NIID 2018a, b; Ministry of Health, Labour and Welfare of Japan 2018). These findings suggest that a substantial number of Japanese individuals remained susceptible to measles after the reported date of elimination; hence, the level of herd immunity over 95% was required to eradicate measles. Rubella infections still occur, including a 2013 nationwide rubella epidemic. Of the 5,442 cases

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reported during the epidemic, 89% occurred in individuals aged > 20 years; however, an increase in congenital rubella syndrome (CRS) was also reported (CDC, Centers for Disease Control and Prevention 2013). The Japanese Ministry of Health, Labour and Welfare (2014) indicated that an antibody titer of ≥ 32 IU/mL was sufficient to prevent rubella infection in pregnant women. In 2014, for the protection of medical workers against rubella infection, JSIPC, the Japanese Society for Infection Prevention and Control (2014) recommended either maintaining an antibody titer of ≥ 32 IU/mL or implementing routine two-dose vaccination. To prevent CRS, it is necessary to prevent transmission in nursery schools and kindergartens as well as in medical facilities. It is important to confirm the uptake of two-dose measles-rubella vaccination in college students majoring in childcare. In 2015, for the protection of workers in nursery schools and kindergartens against measles and rubella infection, Ikuei Junior College implemented serological screening and ascertained vaccination history by using maternity passbooks among students majoring in childcare according to recommendation by JSIPS in 2014. In this study, we investigated the immunization history and seroprevalence of IgGs against measles and rubella among Japanese female college students majoring in childcare.

Materials and Methods

A total of 841 Japanese female students (aged 18.5 ± 0.5 years), majoring in childcare at Ikuei Junior College, were serologically screened for IgGs against measles and rubella. These students were born between April 2, 1996 and April 1, 2000 and were contacted, and their vaccination history was ascertained using their maternity passbooks. We analyzed the newly enrolled students at the time of each admission of four successive years. Students with a history of measles or rubella infection were excluded. Students who had not received two measles or rubella vaccine doses were offered vaccination. The study was approved by the Ethics Committee of the Department of Childcare, Ikuei Junior College. There were 15 male students majoring in childcare during 2015 and 2018 in Ikuei Junior College. All of 15 male students were serologically screened and ascertained vaccination history by using their maternity passbooks. However, we excluded all of 15 male students because of 2 reasons, 1) small number of male students, and 2) greater acceptance of rubella immunization by women in view of the potential risk of CRS has been reported to create a difference in seroprevalence of rubella between men and women (Wicker et al. 2007).

Titers of anti-measles IgGs were measured via the particle agglutination assay using the SERODIA[®] MEASLES kit (Fuji Rebio, Tokyo, Japan) and titers of anti-rubella IgGs were measured via the hemagglutination inhibition (HI) assay using the rubella virus HI "SEIKEN" kit (Denka Seiken, Tokyo, Japan). Both assays were performed according to the manufacturer's instructions. The JSIPC (2014) recommends that antibody titers against measles and rubella should be within the seroprotective range for healthcare workers, including students and volunteers who may have contact with patients. The 841 subjects were classified as seronegative, seropositive, or seroprotected according to the JSIPC criteria. The association

between the number of vaccine doses received and antibody titers was retrospectively analyzed. The JSIPC (2014) recommended seroprotective levels were ≥ 256 IU/mL for measles IgG and ≥ 32 IU/mL for rubella IgG. The Japanese Ministry of Health, Labour and Welfare (2014) recommends a rubella-specific IgG level of ≥ 32 IU/mL to prevent rubella infection. This guideline was intended to prevent rubella infection in pregnant women. To protect pregnant women from rubella infection, immunization of children and workers in kindergartens and nursery schools is necessary. Therefore, seroprotection was defined as a measles-specific IgG titer of ≥ 256 IU/mL and a rubella-specific IgG titer of ≥ 32 IU/mL, seropositivity as a measles-specific IgG titer of 16-128 IU/mL and rubella-specific IgG titer of 8-16 IU/mL, and seronegativity as a measles-specific IgG titer of < 16 IU/mL and a rubella-specific IgG titer of < 8 IU/mL.

Tukey's analysis was performed to compare IgG titers between subjects receiving a single vaccination dose versus two doses, those receiving a single dose versus three doses, and those receiving two versus three doses. Spearman correlation coefficients were calculated to investigate the relationship between measles and rubella antibody titers. Statistical significance was set at $p < 0.05$.

Results

Number of vaccine doses received

The number of measles and rubella vaccine doses received among the 841 female students majoring in childcare as well as their serological status are shown in Fig. 1 and Table 1. Only two students had not been vaccinated against rubella (Fig. 1, Tables 1 and 2). Among the 839 students who had been vaccinated against rubella, 120 (14.3%) received a single dose and 719 (85.7%) received two doses. Among the 120 students who had received a single dose of rubella vaccine, 84 had been vaccinated during childhood (1 year of age) and 36 had been vaccinated in their early teens (12-13 years of age) (Tables 1 and 2). Among the 841 students who had been vaccinated against measles, 103 (12.2%) had received a single dose and 738 (87.8%) had received two doses. Among the 103 students who had received a single dose of measles vaccine, 88 had been vaccinated during childhood and 15 had been vaccinated in their early teens (Tables 1 and 3).

Seroprevalence of rubella-specific IgG

The 839 students who had received one or two doses of rubella vaccine were classified into three groups based on their rubella IgG titers according to the recommendations of the Japanese government and JSIPC (JSIPC 2014; Ministry of Health, Labour and Welfare of Japan 2014). Among the 839 students, 371 (44.2%) were seropositive and 462 (55.1%) were seroprotected (Fig. 1A, Tables 1 and 2). The proportion of students receiving a single dose of rubella vaccine in their early teens who were seroprotected was significantly higher than that of students receiving a single dose of vaccine in childhood or students receiving two vaccine doses (Fig. 2A). Six students who had received one or two doses of rubella vaccine were classified as seronegative (Tables 1 and 2). Antibody titers against rubella were undetectable in two unvaccinated students, and these

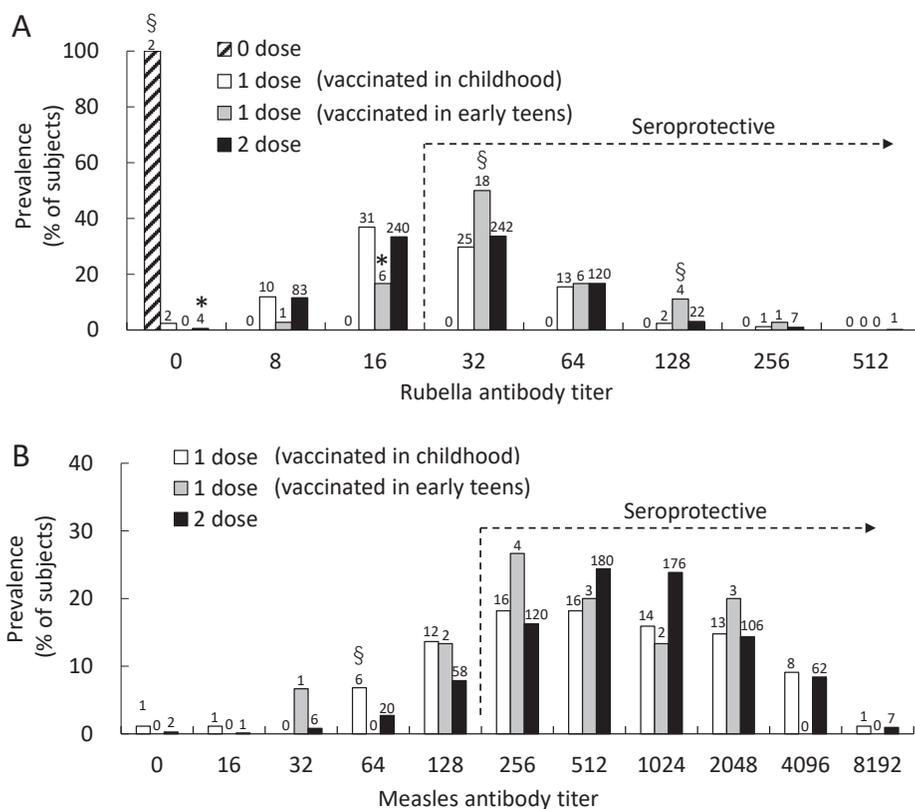


Fig. 1. Number of vaccine doses received and antibody titers against rubella and measles among 841 Japanese female college students.

A: Comparison of rubella-specific antibody titers (IU/mL) among students receiving a single dose of vaccine during childhood, a single dose in their early teens, or two doses. B: Comparison of measles-specific antibody titers (IU/mL) among students receiving a single dose of vaccine during childhood, a single dose in their early teens, or two doses. Number above the column were students' number of each column. Rubella- and measles-specific antibody titers among groups were compared using Tukey's analysis. Statistical significance was assumed at $p < 0.05$.

§significantly higher than other groups.

*significantly lower than other groups.

Childhood, aged 1 year; early teens, aged 13 years.

students were presumably susceptible to rubella although they remained seronegative (Fig. 1A, Fig. 2A, Tables 1 and 2). The proportion of students receiving two doses of rubella vaccine was highest in 2018 and lowest in 2016 (Table 2). The proportion of students who were seroprotected against rubella was highest in 2017 and lowest in 2018 (Table 2).

Seroprevalence of measles-specific IgG

The 841 students were classified into three groups based on their measles IgG titers according to the JSIPC recommendations (Fig. 1B, Fig. 2B, Tables 1 and 3). Among the 841 students, 107 (12.7%) were seropositive and 731 (86.9%) were seroprotected (Tables 1 and 3). The proportion of students receiving two doses of measles vaccine who were seroprotected was significantly higher than that of students receiving a single dose (Fig. 2B). Additionally, the proportion of students receiving a single dose of measles vaccine in childhood who were seroprotected was significantly lower than that of students receiving

two doses or one dose in their early teens (Fig. 2B). Three students who had received one or two doses of measles vaccine remained seronegative. The proportion of students receiving two dose of measles vaccine was highest in 2018 and lowest in 2016 (Table 3). The proportion of students who were seroprotected against measles was highest in 2018 and lowest in 2017 (Table 3).

Discussion

In this study, we found that > 92% of female college students who majored in childcare received two-dose measles-rubella vaccination as of April 2018 and that the number of vaccine doses received by students correlated with IgG titers against measles, but not against rubella, at the time of college admission. In 2015, WHO confirmed that endemic transmission of measles had been eliminated in Japan (WHO 2015). To eliminate measles and rubella, Japan implemented a two-dose measles and rubella vaccination schedule for children in 2006 and a 5-year vaccination catch-up campaign for both measles and rubella from

Table 1. Number of vaccine doses received and serological status against measles and rubella.

Pathogen	Antibody titer	0 dose	1 dose		2 doses	Total
		n (%)	Childhood n (%)	Early teens n (%)	n (%)	n (%)
Rubella	Seronegative (< 8)	2 (100 %)	2 (2.4 %)	0 (0 %)	4 (0.6 %)	8 (1.0 %)
	Seropositive ($\geq 8, < 32$)	0 (0 %)	41 (48.8 %)	7 (19.4 %)	323 (44.9 %)	371 (44.1 %)
	Seroprotective (≥ 32)	0 (0 %)	41 (48.8 %)	29 (80.6 %)	392 (54.5 %)	462 (54.9 %)
	Total	2 (100 %)	84 (100 %)	36 (100 %)	719 (100 %)	841
Measles	Seronegative (< 16)	0 (0 %)	1 (1.1 %)	0 (0 %)	2 (0.3 %)	3 (0.4 %)
	Seropositive ($\geq 16, < 256$)	0 (0 %)	19 (21.6 %)	3 (20.0 %)	85 (11.5 %)	107 (12.7 %)
	Seroprotective (≥ 256)	0 (0 %)	68 (77.3 %)	12 (80.0 %)	651 (88.2 %)	731 (86.9 %)
	Total	0 (0 %)	88 (100 %)	15 (100 %)	738 (100 %)	841

Seroprotection was defined as a rubella-specific IgG titer of ≥ 32 IU/mL and a measles-specific IgG titer of ≥ 256 IU/mL, seropositivity as a rubella-specific IgG titer of 8-16 IU/mL and a measles-specific IgG titer of 16-128 IU/mL, and seronegativity as a rubella-specific IgG titer of < 8 IU/mL and a measles-specific IgG titer of < 16 IU/mL. These classification was performed according to the guideline provided by the Japanese Society for Infection Prevention and Control (JSIPC).

Childhood, the subject who received one-dose vaccination aged 1 year; Early teens, the subject who received one-dose vaccination aged 13 years.

Table 2. Number of vaccine doses received and serological status against rubella in 2015, 2016, 2017, and 2018.

Year	0 dose	1 dose		2 doses	Seronegative	Seropositive	Seroprotective
	number (%)	Childhood number (%)	Early teens number (%)	number (%)	number (%)	number (%)	number (%)
2015	2 (1.1 %)	22 (11.6 %)	11 (5.8 %)	154 (81.5 %)	3 (1.6 %)	75 (39.7 %)	111 (58.7 %)
2016	0 (0.0%)	34 (16.4%)	12 (5.8%)	161 (77.8 %)	3 (1.5 %)	100 (48.3 %)	104 (50.2 %)
2017	0 (0.0%)	17 (7.11%)	8 (3.4%)	214 (89.5 %)	1 (0.4 %)	94 (39.3 %)	144 (60.3 %)
2018	0 (0.0%)	12 (5.8%)	4 (1.9%)	190 (92.2 %)	1 (0.5 %)	102 (49.5 %)	103 (50.0 %)

Serological status against rubella among 841 students were screened by the hemagglutination inhibition assay. One hundred eighty nine students were serologically screened in 2015, 207 students were screened in 2016, 239 students were screened in 2017 and 206 students were screened in 2018. Serological status against rubella among 841 students were classified as seronegative, seropositive and seroprotective according to the guideline provided by the Japanese Society for Infection Prevention and Control.

2008 to 2012 (Kimura et al. 2013; IDSC, NIID 2015). In 2013, a nationwide rubella epidemic occurred in Japan, and an increase in CRS was reported (CDC 2013). It is critical to prevent rubella infection in kindergartens and nursery schools as well as in medical facilities. The JSIPC has provided vaccination guidelines for preventing outbreaks of vaccine-preventable diseases in medical facilities. The

JSIPC guidelines recommend two-dose vaccination as well as classification of antibody titers as seronegative, seropositive or seroprotective (JSIPC 2014).

Seroprevalence of rubella-specific IgG

Here, the proportion of subjects who were seropositive or seroprotected against rubella was 99.0%. However, the

Table 3. Number of vaccine doses received and serological status against measles in 2015, 2016, 2017, and 2018.

Year	0 dose	1 dose		2 doses	Seronegative	Seropositive	Seroprotective
	number (%)	Childhood number (%)	Early teens number (%)	number (%)	number (%)	number (%)	number (%)
2015	0 (0.0 %)	25 (13.2 %)	0 (0.0 %)	164 (86.8 %)	0 (0.0 %)	27 (14.3 %)	162 (85.7 %)
2016	0 (0.0 %)	34 (16.4 %)	8 (3.9 %)	165 (79.7 %)	2 (1.0 %)	26 (12.6 %)	179 (86.5 %)
2017	0 (0.0 %)	17 (7.1 %)	5 (2.1 %)	217 (90.8 %)	1 (0.4 %)	37 (15.5 %)	201 (84.1 %)
2018	0 (0.0 %)	12 (5.8 %)	2 (1.0 %)	192 (93.2 %)	0 (0.0 %)	17 (8.3 %)	189 (91.8 %)

Serological status against measles among 841 students were screened by the particle agglutination assay. One hundred eighty nine students were serologically screened in 2015, 207 students were screened in 2016, 239 students were screened in 2017 and 206 students were screened in 2018. Serological status against measles among 841 students were classified as seronegative, seropositive and seroprotective according to the guideline provided by the Japanese Society for Infection Prevention and Control.

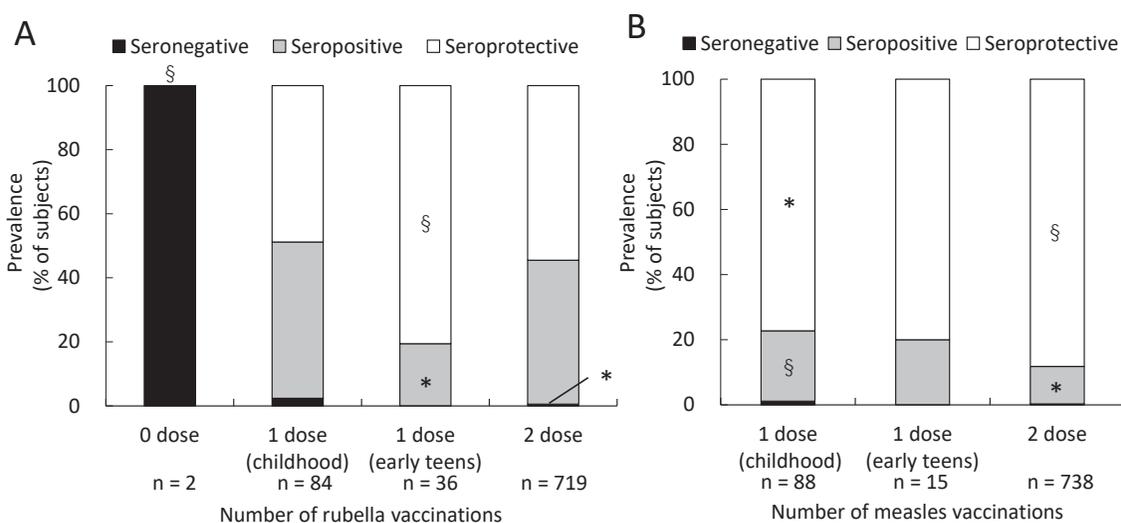


Fig. 2. Proportion of 841 Japanese female college students with seronegative, seropositive and seroprotective antibody titers against rubella and measles.

Panel A represents antibody titers against rubella, and panel B represents antibody titers against measles. Students were stratified into four groups depending on the number of vaccine doses received: (i) a single dose during childhood, (ii) a single dose in their early teens, (iii) two doses, and (iv) unvaccinated. The proportions of seronegative, seropositive or seroprotected students in each stratum were compared by Tukey's analysis. Statistical significance was assumed at $p < 0.05$.

§significantly higher than other groups.

*significantly lower than other groups.

Childhood, aged 1 year; early teens, aged 13 years.

proportion of seroprotected individuals against rubella, as defined by the JSIPC and the Japanese government, was much lower (54.9%; Tables 1 and 2). The proportion of students receiving one dose of rubella vaccine in their early teens who were seroprotected was significantly higher than that of students receiving two doses of vaccine or one dose during childhood (Fig. 2A). The proportions of students receiving a single dose of rubella vaccine in childhood or

two doses of vaccine who were seroprotected were similar (Fig. 2A). Similar results were reported by a national surveillance study of rubella in 2010 (IDSC, NIID 2011). These findings could be explained in three ways: 1. antibody titer measured by HI method is nonconsecutive, 2. pre-existing antibodies against rubella prevented an increase in antibody titer after booster vaccination (Tischer and Gerike 2000), 3. there was a significant association between

seronegativity and the number of years elapsed since vaccination (Poethko-Müller and Mankertz 2012). In support of the last hypothesis, waning of rubella antibody titers has been reported (Pebody et al. 2002; Amanna et al. 2007; Poethko-Müller and Mankertz 2012). The half-life of serum IgG was found to be over 85 years for rubella (Amanna et al. 2007). In the present study, the subjects were screened within 5-6 years of their last vaccination. In the case of immunization against rubella, the number of vaccine doses received was not correlated with antibody titer. Recent reports showed that vaccine efficacy is higher after two doses of measles, mumps, and rubella (MMR) vaccination than after a single dose (Hennessey et al. 1999; Paunio et al. 1999; Vitek et al. 1999), despite that fact that population antibody titers returned to pre-second dose levels 3 years after administration of the second dose. These observations suggested that low antibody titers in highly-vaccinated individuals are not necessarily indicative of a lack of clinical protection (Pebody et al. 2002). Together, these data support the recommendation of the Japanese government and JSPIC that routine two-dose vaccination without measuring antibody titer against rubella should be implemented to protect against infection.

Seroprevalence of measles-specific IgG

Among the 738 students who had received two doses of measles vaccine, 651 (88.2%) were seroprotected. The proportion of students who had received two vaccine doses who were seroprotected against measles was significantly higher than that of students who received a single vaccine dose (Fig. 2B). These results may relate to the reported half-life of over 300 years for measles-specific serum IgG (Amanna et al. 2007). In the case of immunization against measles, the number of vaccination doses was proportional to antibody titer.

To prevent measles and rubella infection, JSIPC (2014) recommended either maintaining antibody titer at or above the seroprotective level or the implementation of two-dose vaccination schedules. The proportion of students receiving two doses of measles and rubella vaccines increased from 2015 to 2018, except in 2016. In this study, the prevalence of students who had not received two doses of measles and rubella vaccines was over 10% at the time of college admission. This result suggests the importance of targeted immunization of Japanese students majoring in childcare prior to their clinical training to prevent the transmission of serious diseases among children and nursery school personnel.

In conclusion, the proportion of students receiving two doses of measles and rubella vaccines was increasing over the period surveyed (2015 to 2018); however, a substantial number of students remained susceptible to these diseases at the time of college admission. We propose the implementation of targeted immunization of Japanese college students majoring in childcare, as well as medical students, prior to their clinical training to prevent the transmission of

these serious diseases among children and nursery school personnel.

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Conflict of Interest

The authors declare no conflict of interest.

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